REMARKS

In response to the Final Office Action mailed June 17, 2008, Applicants have the following remarks:

- All claims are now directed to a method of detecting a target nucleic acid in a ***living cell.*** This is not taught in Landegren *et al.* (US2002/0064779), which teaches nucleic-acid probe driven *nucleic acid complementation* which can only be performed in a **ruptured** cell but not in a living cell. The Landegren system also requires either a high degree of nucleic acid complementation or secondary analysis step to detect nucleic acid complementation. Michnick's (US 6,270,964) teaches a protein-probe driven *protein complementation*. The binding properties of protein probe to drive complementation is very different from the binding properties of a nucleic acid probe. A skilled artisan would not be motivated to substitute the complementary nucleic acids in the method of Landegren with the polypeptide complex of Michnick, or have a reasonable expectation that a nucleic acid probe-driven method would work in a living cell. Applicants respectfully submit amendments to Claims 1 and 21 obviates the §103 obviousness rejections of Landegren *et al.* in view of Michnick *et al.* or in further view of Sodroski *et al.* (US 5,654,195).
- Claim 20 is directed to a clarify that the "target *nucleic acid sequence* forms part of the triplex" complex. This is not taught by Stefano *et al.,* (US 6,287,772) which teaches three separate *probes* from a triplex; *two nucleic acid probes* which bind to separate target sequences and a *third nucleic acid probe* which binds with the two nucleic acid probes. Applicants respectfully submit amendment to Claims 20 obviates the §103 obviousness rejections of Stefano *et al.,* in view of Landegren *et al* and Michnick *et al.*

Summary of Amendments to the claims:

- Claim 1 and 21 have been amended to be directed to nucleic acid probe driven protein complementation "***in a living cell***" (i.e. in an intact cell). Support for these amendments are in paragraphs [028] and [046] of the published application.

- New dependent Claims 25-28 claim a living cell is *in vivo* or *in vitro*. These amendments are supported by paragraphs [018], [028], [039], [046] and [081] of the published application.

- Claim 14 has been amended to be properly dependent on Claim 1. This is supported throughout the specification, for example paragraphs [022], [044], [0056] to [0059] of the published application.

- Claim 20 has been amended to clarify that the "*target nucleic acid sequence forms part of the triplex*" complex. This is supported by paragraphs [026] and [057] of the published application.

Accordingly, these amendments do not constitute new subject matter and Applicants respectfully request their entry.

Detailed Response

Claim rejections under 35 U.S.C. §112, second paragraph (written description):

Claim 14 is rejected under 35 U.S.C. §112, second paragraph for being indefinite for failing to particularly point out the subject matter, as it depends on canceled claim 13. Applicants respectfully submit amendments to claim 14 has obviated this rejection. This amendment is supported throughout the specification, in particular in paragraphs [022], [044], [0056] to [0059] of the published application.

Claim rejections under 35 U.S.C. §103 (obviousness):

Claims 1-3, 5-12, 14, 16, 17, 19 and 21-23 are rejected under 35 U.S.C. §103 as being obvious by Landegren (US 2002/0064779) in view of Michnick (US 6,270,964). The Examiner has also rejected Claims 4 and 24 as being as being obvious by Landegren et al., (US 2002/0064779) in view of Michnick (US 6,270,964) and in further view of *Sodroski* (US 5,654,195), and Claim 20 in view of Landegren et al., (US 2002/0064779), Michnick (US 6,270,964) and Stefano (6,287,772).

The Examiner has argued that in Landegren teaches at paragraphs [0007] and [0010] the use of a nucleic acid binding moiety to detect a nucleic acid target analyte to bring about nucleic acid complementation. The Examiner has acknowledged that neither Landegren or Michnick teach the present invention, but contends that Landegren teaches protein or nucleic acid driven *complementation of nucleic acids*. Michnick teaches a method of protein probe driven *protein complementation* to detect *proteins*. The Examiner argues that one of ordinary skill in the art would be motivated to **substitute** the complementary nucleic acids in the method of Landegren with the polypeptide complex of Michnick, and that it would be reasonably expected to use protein complementation as an alternative detection means to nucleic acid complementation.

However, the Examiner has ignored essential differences between these references and the present invention. Significantly, Landegren's method cannot be used in living cell. It can only be performed *in vitro* **in ruptured cells**. Further, Michnick's method is **protein-probe driven** protein complementation. Landergen's system is designed to function in ruptured cells which is a fundamentally different environment to working in intact cells, so it would not be reasonable to expect to use such a system in a living cell. There is also no reasonable expectation of success should one of ordinary skill in the art substitute the *in vitro* nucleic acid complementation detection method of Landergen with the polypeptide-complementation detection method of Michnick.

Landegren's teaches a nucleic-acid probe driven nucleic acid complementation to detect *nucleic acids*. However, this method requires either a high degree of nucleic acid complementation (i.e., a lot of the target), or a secondary analysis step such as amplification (please see [0014]) to detect the nucleic acid complementation. This is not a difficult step in a ruptured cell, but it can ***not*** be performed in an *intact living cell* (i.e. *in vivo*) or in *real time*. This requirement for a large amount of the nucleic acid target or an amplification step also shows why one would not substitute Michnick's protein-probe driven method by a nucleic acid-probe driven method.

The binding properties of a *protein probe* to drive complementation is very different from the binding properties of *nucleic acid probe*. Landegren does not teach either the specificity of the nucleic acid probe required drive efficient nucleic acid complementation, nor of the spacing of the nucleic acid probes on the target nucleic acid sequence for nucleic acid complementation. Furthermore, Landegren does not teach methods to enable the nucleic acid probes to detect point mutations or variations in the target nucleic acid sequence. Thus the skilled artisan would not expect that one could use a nucleic acid probe-driven method in a living cell because one would expect that one would need large amounts of nucleic acid or to use a separate amplification step. The only way one would know that one does not need large amounts of the nucleic acid target or a separate amplification step is by hindsight based upon the present disclosure.

Accordingly, Applicants submit that the Examiner is clearly wrong to suggest that one of ordinary skill in the art would expect to be successful in substituting the complementary nucleic acids in the method of Landegren with the polypeptide complex of Michnick to result in the present invention for detection of nucleic acids using a combination of nucleic acid probe driven protein complementation.

Applicants have made this explicit by the amendments to Claims 1 and 21 to nucleic acid probe driven protein complementation "***in a living cell***" (i.e. in an intact cell). These amendments are supported by paragraphs [028], [039], and [046] of the published application and do not introduce new subject matter. Accordingly, Applicants submit that amendments to claims 1 and 21 have obviated the rejections and respectfully request the rejection be withdrawn.

For the reasons stated above, Applicants also disagree that Claims 4 and 24 are obvious by Landegren *et al.,* (US 2002/0064779) in view of Michnick (US 6,270,964) and in further view of *Sodroski* (US 5,654,195). Further, Sodroski teaches antibodies against discontinuous epitopes on a single molecule, i.e. a HIV-1 protein (see column 12, lines 41-43), but does not specifically teach a method of using an antibody to detect a discontinuous epitopes from two *separate proteins* or from separate polypeptides which have come together by protein complementation. Thus, Applicants respectfully request the rejection be withdrawn.

Claim 18 is also rejected under 35 U.S.C. §103 for being obvious over Landegren *et al.,* in view of Michnick and in further view of *Lizardi* (US 5,854, 033). The Examiner argues that one of ordinary skill in the art would have been motivated to use rolling circle amplification (RCA) to provide target nucleic acids in the method of Landegren as modified by Michnick because Lizardi disclosed RCA was a good means of providing amplified levels of nucleic acids. For the reasons stated above, and contrary to the present invention, Landegren's method requires a high degree of nucleic acid complementation (i.e. a lot of target), thus the only way one would know to use RCA is hindsight based upon the present invention. Thus Applicants disagree that Claims 18 is obvious by Landegren *et al.,* in view of Michnick and in further view of Lizardi (US 5,854, 033) and respectfully request the rejection be withdrawn.

Applicants also respectfully disagree with the Examiner that Claim 20 is obvious in view of Landegren, Michnick and Stefano (US 6,287,772). Applicants point out to the Examiner that Stefano teaches a triplex of PNA (peptide nucleic acid) which is a pseudopeptide. A pseudopeptide is not equivalent to duplex nucleic acid probes, as stated in column 2, line 16-17 of Stefano. Furthermore, Applicants point out that in Figure 1, Stefano teaches detection of a target nucleic acid by the hybridization of two PNA probes (X and Y) to *different target sequences* on the target nucleic acid. The non-bound arm segment of two PNA probes interact with a *third PNA probe* (Z) to form a triplex [see also col 8, (lines 4-6) and col 9 (lines 7)]. Importantly and unlike the present invention, Stefano does not teach the binding of the two PNA probes to the ***same site*** on the target nucleic acid to form a triplex, nor does it teach the target nucleic acid being one of the three nucleic acid sequences making up the triplex. Accordingly, the combination of Landegren, Michnick and Stefano does not teach each and every limitation of the present invention. However, in order to expedite prosecution, Applicants have amended Claim 20 to clarify that the target nucleic acid is part of the triplex complex, and that amended Claim 20 which is supported by paragraphs [026] and [057] of the published application has obviated the rejection and respectfully request the rejection be withdrawn.

In the event that there are any questions relating to this Amendment, or to the application in general, it is kindly requested that the Examiner contact the undersigned attorney concerning the same to expedite the prosecution of this application.

In view of the foregoing, Applicants respectfully submit that all claims are in condition for allowance. Early and favorable action of the subject application on the merits is respectfully requested.

FEE AUTHORIZATION

The Examiner is authorized to charge any fee deficiencies or credit any overpayments associated with this submission to the Nixon Peabody LLP Deposit Account No. 50-0850. In the event that there are any questions concerning this response, or the application in general, the Examiner is respectfully urged to telephone the undersigned so that prosecution of the application can be expedited.

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Respectfully Submitted,

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